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Directional effect of inactivation of the nucleus of the optic tract on optokinetic nystagmus in the cat

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Abstract

The goal of the present investigation was to elucidate the role of the nucleus of the optic tract and the dorsal terminal nucleus of the accessory optic system (NOT-DTN) for slow eye movements other than horizontal. Retinal slip neurons in the NOT-DTN in the awake behaving cat respond direction selectively to the ipsiversive component of horizontal and oblique image motion. They are, however, influenced neither by pure vertical stimulus movement nor by eye movements in the dark. Electrical stimulation of the NOT-DTN leads to pure horizontal optokinetic nystagmus with ipsiversive slow phases and does not influence vertical eye position. In addition, unilateral reversible inactivation of the NOT-DTN with muscimol elicits spontaneous contraversive horizontal nystagmus without vertical component. During oblique optokinetic stimulation, the ipsiversive OKN component is significantly decreased in all directions. After bilateral NOT-DTN inactivation, OKN can only be elicited in a narrow range of upward directions. These data indicate that the NOT-DTN is the only source to drive the horizontal component of OKN. © 2001 Elsevier Science Ltd. All rights reserved.

Keywords: Accessory optic system; Electrical stimulation; Reversible inactivation; Muscimol-optokinetic reflex

1. Introduction

A prerequisite for visual analysis of an object is its temporary stabilization on the retina. This requires minimization of image slip for which most seeing animals have evolved stabilizing reflexes for the eyes in the head and the head on the trunk, as the optokinetic reflex (OKR) and the vestibulo-ocular reflex. In mammals, the OKR is subserved by the pretectal nucleus of the optic tract (NOT) and the terminal nuclei of the accessory optic system (AOS) (for a review, see Simpson, Giolli, & Blanks, 1988). The AOS consists of three nuclei located at the surface of the midbrain: the dorsal terminal nucleus (DTN), the lateral terminal nucleus (LTN), and the medial terminal nucleus (MTN).

In the NOT of the awake cat, neurons with different response properties have been described and related to eye movements (Schweigart & Hoffmann, 1992; Schmidt, 1996). One class of neurons termed 'retinal slip neurons' can be characterized by their direction selective responses to slow horizontal movements of a retinal image (Ballas & Hoffmann, 1985). These neurons respond with a strong directional selectivity to the ipsiversive component of stimulus movements, i.e. leftward components in the left NOT, rightward components in the right NOT. Similar neurons can be found in the DTN, and we consider NOT and DTN as a functional unit (NOT-DTN). Retinal slip neurons are involved in the control of slow-phase horizontal eye movements during OKR (Simpson et al., 1988). This has been studied directly so far by recording neurons and eye-movements simultaneously in monkeys only (Mustari & Fuchs, 1990; Ilg & Hoffmann, 1991). In addition, retinal slip neurons have been identified under anaesthesia in a variety of mammalian species (rat: e.g. Cazin, Precht, & Lannou, 1980; rabbit: Collewijn, 1975b; ferret: Klauer, Sengpiel, & Hoffmann, 1990; cat: Hoffmann & Schoppmann, 1981; Grasse & Cynader, 1984; opossum: Volchan, et al., 1989; Ibbotson, Mark, & Maddess, 1994).

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In a model proposed by Hoffmann (1982) the generation of the horizontal slow eye-movements stabilizing gaze (like during optokinetic nystagmus (OKN)) can be explained by the activity difference between retinal slip neurons in the left and right NOT-DTN. The activity difference between the two nuclei evolves by an increase in the activity in one nucleus and a decrease in activity in the opposite nucleus caused by the same stimulus direction. In darkness, when NOT-DTN cells are not excited by any stimulus, there should be no activity difference and, consequently, no nystagmic eye movements. This is demonstrated for a cat during normal looking in Fig. 1. Electrical stimulation or lesioning of one NOT-DTN should result in a destabilization of the system, and nystagmic movements towards the side of the active NOT-DTN should be generated. Indeed, this could be shown in rabbit (Collewijn 1975a), cat (Precht & Strata, 1980) and monkey (Kato et al., 1986; Kato, Harada, Hasegawa, & Igarashi, 1988; Schiff, Cohen, Büttner-Enever, & Matsuo, 1990; Ilg, Bremmer, & Hoffmann, 1993; Yakushin et al., 2000). Electrical stimulation in the NOT-DTN resulted in a nystagmus with slow phases towards the stimulated side, and an electrolytic lesion resulted in a reduction or inability to perform horizontal OKN towards the side of the lesion.

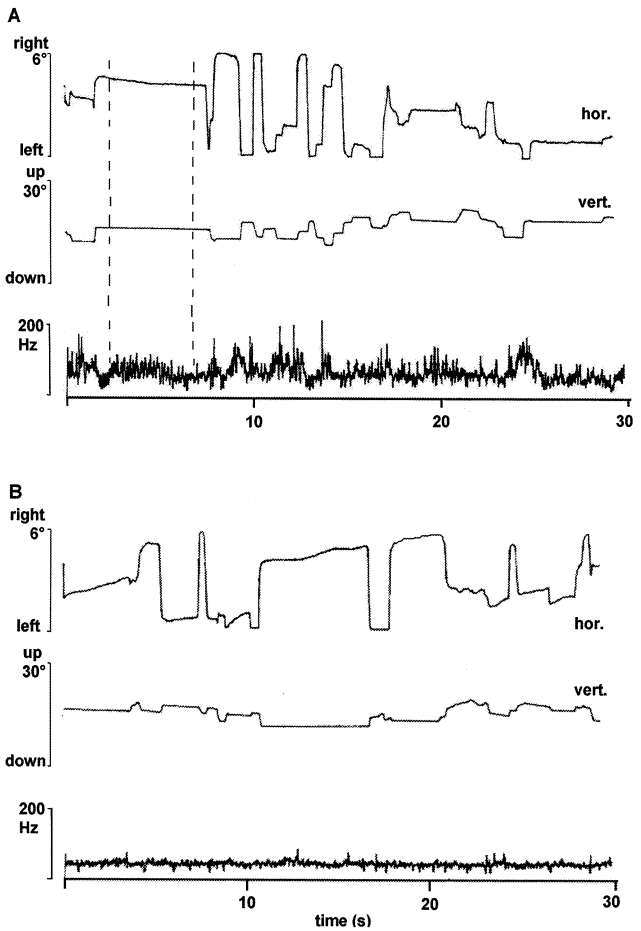

Fig. 1. Response properties of a representative neuron in the right NOT-DTN during spontaneous eye movements in the light while the cat was looking at a stationary Julesz pattern (A) and during darkness (B). The upper and middle traces give the horizontal and vertical eye position, respectively. Lower traces depict the neuronal response during 30 s recording time. Calibration bars represent 6° for horizontal and 30° for vertical and 200 Hz firing rate. Upward deflections of the eye position traces correspond to rightward and upward eye movements. Vertical broken lines in (A) give the segment that is referred to in the text.

Cats, monkeys, and humans have retinae possessing an area centralis or fovea, and their optic nerves project to both sides of the brain. This seems to be the prerequisite for symmetrical horizontal monocular OKN. Proper retinal image stabilization, however, requires OKN in all directions. Vertical OKN seems to be controlled by the MTN and LTN (Simpson et al., 1988). In foveate mammals, vertical OKN was shown to be asymmetrical with different gain for up- and downward movement (cat: Grasse & Cynader, 1988; monkey: Matsuo & Cohen, 1984; human: van den Berg & Collewijn, 1988; Murasugi & Howard, 1989; Ogino, Kato, Sakuma, Takahashi, & Takeyama, 1996).

From this knowledge, the question arises whether there is an influence of temporary inactivation of the NOT-DTN onto OKN in all directions. The effects of a temporary inactivation of both NOT-DTNs onto OKN is also unknown. Thus, we tested a prediction of oculomotor behaviour that resulted from unilateral NOT-DTN lesions by temporarily inactivating the NOT-DTNs on both sides. We compared the effects of transitory lesions of the NOT-DTN induced by the GABA agonist muscimol on nystagmic eye movements with the electrophysiological data from single cell recordings of NOT-DTN neurons in the awake cat. A preliminary report on the present findings has been published in abstract form (Fischer, Schmidt, & Hoffmann, 1997).

2. Methods

2.1. Surgery

All experiments were approved by the local ethics committee and were carried out in accordance with the European Communities Council Directive of 24 November 1986 (S6 609 EEC) and NIH guidelines for care and use of animals for experimental procedures. Adult cats of both sexes that had been purpose-bred in our own animal facility were initially anaesthetized with a mixture of ketamine (20 mg/kg body weight) and thiazinhydrochloride (Rompun®1 mg/kg). The animals were intubated through the mouth, and an intravenous catheter was inserted into the forearm vein. Then, they were placed in a stereotaxic frame and artificially ventilated with a 3:1 mixture of nitrous oxide and carbogen

(95% O_2, 5% CO_2). Anaesthesia was maintained by adding 0.2–0.4% halothane to the gas mixture. After additional local anaesthesia with bupivacain hydrochloride (Bupivacain®) or prilocainhydrochloride (Xylonest®), the skin overlying the skull was cut, and a scleral search coil (Judge, Richmond, & Chu, 1980) and head holder were implanted in all animals under aseptic conditions. In addition, in cats participating in recording experiments ($n = 2$), a recording chamber was implanted to allow access to the midbrain and pretectum. In animals for inactivation experiments ($n = 3$) a 26-gauge stainless steel guide tube aimed 2 mm above the region of the pretectum was implanted. In order to facilitate the localization of the guide tube prior to its positioning, the NOT-DTN was localized with electrophysiological recordings. When the typical increase of cell activity was found by stimulating with horizontal temporonasal pattern movement, the recording electrode was retracted, and the guide tube was inserted at the same place with the tip 2 mm above the measured cell activity. Between experiments, the clearance of the guide tube was protected by a stainless steel stylet and closed by a screw-top cap. During the whole surgical procedure, heart rate, body temperature, and endtidal CO_2 were monitored and kept at physiological levels. After complete recovery, the animals were returned to their animal quarter. They were treated with antibiotics and analgetics for 5–7 days after surgery.

2.2. Recording and electrical stimulation

In alert cats, single units in the NOT-DTN were recorded extracellularly with tungsten in glass microelectrodes (1–5 MΩ at 1 kHz), conventionally amplified, high-pass filtered, and fed into the lab-interface of a PDP 11/34 computer for data storage and analysis. The NOT-DTN was localized by aiming the electrodes stereotaxically at the superior colliculus as a guiding structure. For electrical stimulation electric pulses (width: 1 ms, frequency: 60 Hz, typical amplitude: 0.1 mA) were delivered through the recording electrode.

Horizontal and vertical eye movements were monitored using the phase-detection principle in a magnetic field (Kasper & Hess, 1991). Eye movements were measured relative to the stationary head centred in the magnetic field. During the recording sessions, animals were comfortably placed in a plastic box, cushioned with towels to confine body movements, and their heads fixed to the box by a plastic head holder. The cats exhibited no evidence of discomfort and were kept in the box for as long as 2 h.

2.3. Injection experiments

Injections were performed with a modified version of the method described by Bracha, Webster, Winters, Irwin, and Bloedel (1994). The GABA agonist muscimol was delivered through a stainless steel needle inserted into the guide tube. A 1 μl calibrated plastic tubing was connected to the injection needle. The drug was applied with a manually driven 10 μl Hamilton syringe. The injected volume was controlled by observing the movement of a small air bubble placed in the plastic tube close to the injection needle. A total volume of 1 μl of muscimol in saline (1 mg/ml) was injected at a rate of 0.2 μl/min. The injection of the drug started 2 mm above the expected location of the NOT-DTN. The application continued on separate days at increasing depths in 0.5 mm increments until the injection site was found at which muscimol elicited spontaneous nystagmus in darkness.

2.4. Visual stimulation

Visual stimulation was provided by moving a large Julesz pattern (square size 1°; light square 43 cd/m^2; dark square 5 cd/m^2; contrast: 0.79), which was projected onto a tangent screen placed 80 cm in front of the cat so that a 95° by 95° visual field was stimulated. A computer controlled stimulus movements. During recording experiments, the pattern was moved on a circular path with 20°/s. This stimulus movement was useful to quickly calculate the preferred direction of stimulus movement of a recorded neuron (Hoffmann & Schoppmann, 1981). To elicit optokinetic nystagmus, the pattern was moved at velocities of 5–19.8°/s in one direction, and the movement directions were altered in steps of 22.5°. All OKN measurements were done binocularly. To avoid the fast rise and slow build-up component (Maioli & Precht, 1984), and to measure only the steady state of slow-phase eye movements, recordings always started 10 s after the beginning of the optokinetic stimulation.

2.5. Histology

At the end of a successful penetration, microlesions were placed at the recording sites of NOT-DTN units by passing 5 μA (electrode tip positive) for 5 s. At the end of the experiments, the animals were given an overdose of pentobarbital. After respiratory block and complete lack of reflexes, the animals were perfused transcardially with saline and 10% phosphate-buffered formalin. After cryoprotection in 10%, 20%, and 30% sucrose, 50 μm thick frozen sections were cut through the meso-diencephalic area, and stained alternatively for Nissl or Klüver-Barrera for visualization of the electrolytic lesions and the NOT-DTN injection sites.

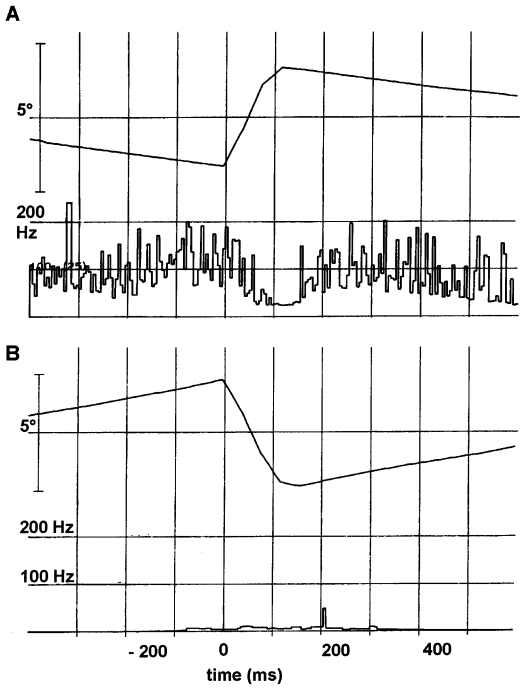

Fig. 2. Response properties of a neuron in the left NOT-DTN to optokinetic stimulation. Responses (lower trace) are aligned to saccade onset at 0 ms (upper trace) and averaged. In (A) responses, to slow-phase eye movements directed to the left and saccades directed to the right are shown, and vice versa in (B). Calibration bars represent 5° and 200 Hz firing rate. Upward deflections of the eye position traces correspond to rightward eye movements.

3. Results

3.1. Recording experiments

In this first paragraph, some of the properties of NOT-DTN cells already described in the anaesthetized preparation will be confirmed in the awake cat to provide some basis for the interpretation of the behavioural deficits due to the inactivation of these cells. The retinal slip cells analyzed here represent a functional subgroup of neurons described in the NOT-DTN of anaesthetized and awake cats (Hoffmann & Schoppmann, 1981; Ballas & Hoffmann, 1985; Schweigart & Hoffmann, 1992; Schmidt, 1996). We mainly report on their direction selectivity and velocity tuning. An example of the response of a cell recorded in the right NOT-DTN during spontaneous eye movements is given in Fig. 1. In the light (Fig. 1A), the activity of the cell plotted as instantaneous frequency was modulated during spontaneous saccades and during periods of imperfect stabilization of gaze. During one of these periods of about 5 s duration indicated by dashed lines, the cell was active at about 100 Hz. In the first two-thirds of this period, the activity of the cell was greater than in the last third, which corresponds to a slight leftward drift of the eye. This drift caused rightward retinal slip that in turn elicited neuronal activity in the right NOT-DTN to stabilize the eye in a horizontal direction. When the eye seemed to be stabilized in the last third of this period, indicated by the absence of horizontal eye drift, the cell activity was reduced.

During darkness (Fig. 1B), the cell's activity was reduced to a constant spontaneous level, and there was no modulation of activity induced by eye movements. Due to the lack of visual input and, as a consequence, no neuronal activity modulation in NOT-DTN to stabilize the horizontal eye position, there are far larger drifts during intersaccadic intervals than in the light. In both light and dark, vertical eye movements seemed to exert no influence on the cell activity.

To further analyze the response properties, the activity of a cell in the left NOT-DTN was recorded during optokinetic stimulation. In Fig. 2, the horizontal eye position and neuronal activity during OKN averaged over 20 saccades are shown. Stimulation into the preferred, i.e. leftward direction (Fig. 2A), elicited a neuronal response of about 100 Hz during leftward slow eye movements. The cell activity was heavily reduced during resetting saccades accelerating retinal slip in the same direction. Stimulation into the non-preferred, i.e. rightward direction (Fig. 2B), totally suppressed the cell activity during rightward slow eye movements. These results show that NOT-DTN neurons responded highly direction-specifically to the remaining retinal slip during the slow phases of OKN. During saccades, the neuronal activity was decreased. This can be explained simply by a non-optimal high velocity during saccadic image shifts. To confirm this claim, the response properties of NOT-DTN cells during optokinetic stimulation were further analyzed with respect to various retinal slip velocities. During constant velocity stimulation, retinal slip was calculated by subtracting eye velocity from external stimulus velocity. The most effective slip velocities were between 5° and 20°/s, comparable to published values in the anaesthetised cat (Hoffmann & Schoppmann, 1981; Grasse & Cynader 1984). High slip velocities, as they occur during saccades, are ineffective in driving NOT-DTN cells.

During single-cell recordings in the right NOT-DTN, electric pulses were delivered through the recording electrode for 10 s while the cat was sitting in the dark (Fig. 3). This elicited a horizontal nystagmus with slow phases directed to the right (upper trace of the eye position trace). The mean of the slow phase eye velocity during the first 10 s was 4.6°/s. In contrast, the vertical eye position was unaffected by the stimulation. This indicates that the electrically elicited activation generated only horizontal eye movements, a finding that helps to explain our results from the inactivation of the NOT-DTN presented in the next section. After 10 s, when the stimulation was switched off, an afternystagmus with slowly decreasing velocity of the slow phases was evident.

3.2. Inactivation experiments

The results from the recording and stimulation experiments in the awake cat show that NOT-DTN cells provide a direction-specific signal about the velocity of retinal slip during OKN. To further investigate the functional role of the NOT-DTN, we temporarily inactivated the pretectum with muscimol injections and recorded changes in eye movement behaviour not only during horizontal but also during oblique and vertical directions of stimulus movement.

3.2.1. Spontaneous nystagmus

Eye-movement behaviour was tested while the cats were sitting in the dark before and soon after application of muscimol into the pretectal region. A nystagmus with slow pases towards the intact side began to develop between 2 and 15 min after injection (termed 'pretectal inactivation'; PTI). These effects were temporary, and full recovery was always observed the next day.

Fig. 4 shows typical spontaneous eye movements in the dark after muscimol application into the left, right, or both pretectal regions. As shown in Fig. 4A, 2 min after injection into the left pretectal region, a spontaneous nystagmus in the horizontal plane developed (upper trace) with linear slow phases directed to the contralateral (right) side. The mean eye velocity of the slow phases was 6.8°/s. The vertical eye movements were not affected by the muscimol injection (lower trace). From a session on the next day, we show in Fig. 4B how, 3 min after injection into the right pretectal region, a horizontal nystagmus developed with slow phases directed to the left at a mean velocity of 8.1°/s. Again, vertical eye movements were unaffected.

In Fig. 4C, both pretectal regions were inactivated. This was not done simultaneously. Instead, first, the left side was injected, and spontaneous nystagmus to the right was observed. Only then was the right side additionally inactivated and horizontal spontaneous nystagmus vanished. This procedure guaranteed that the muscimol was active on both sides. The means of the horizontal slow phase velocities were 2.1°/s to the right and 0.8°/s to the left. This was nearly equal to spontaneous eye movements in the dark during control situations, i.e. 2.0°/s to the right and 0.6°/s to the left. Thus re-establishing the balance between the activity in the left and right NOT-DTN abolishes spontaneous nystagmus.

3.2.2. Controls

During control situations, i.e. prior to the injection experiments, OKN was elicited by moving the stimulus pattern at 12°/s in 12 different directions. The gain of optokinetic slow phases, expressed as the ratio between slow phase eye velocity and stimulus velocity, is displayed in Fig. 5. The radius of the outer circle is equal to a gain of 1, which would indicate that the eye movements could perfectly follow the stimulus velocity. In none of the stimulus directions was a gain of 1 reached, indicating that eye velocity was always lower than stimulus velocity.

During the control situation (open symbols in Fig. 5A, C, E), in all three cats, OKN was approximately symmetrical for horizontal stimulus directions with a gain between 0.5 and 0.8. In contrast, all three cats showed asymmetrical OKN during vertical stimulation. During upward pattern movement, the slow phase gain was equal to horizontal values (Fig. 5 A, C) or even better (E). A downward pattern movement, however, resulted in a dramatic decrease in the slow phase gain.

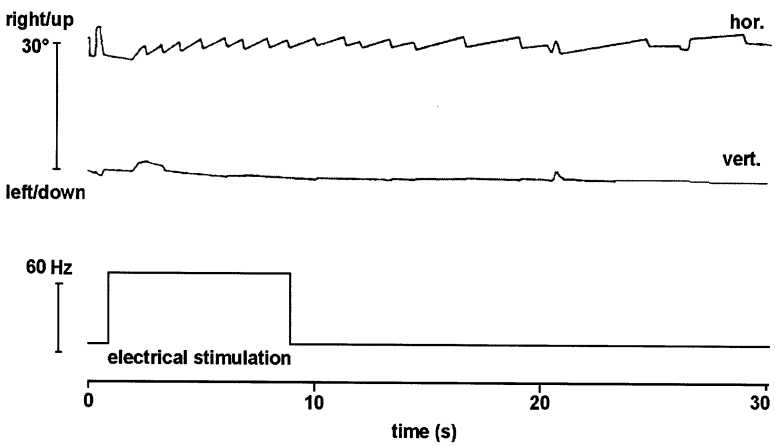

Fig. 3. Electrical stimulation at the recording site in the right NOT-DTN while the cat was sitting in complete darkness. The upper and middle traces show the horizontal and vertical eye movements, respectively. Calibration bars represent 30° and frequency (Hz) of electrical stimulation. Upward deflections of the eye position traces correspond to rightward and upward eye movements. About 1–2 s after the onset of stimulation, an optokinetic nystagmus with rightward slow phases developed. After cessation of stimulation, an optokinetic afternystamus was present.

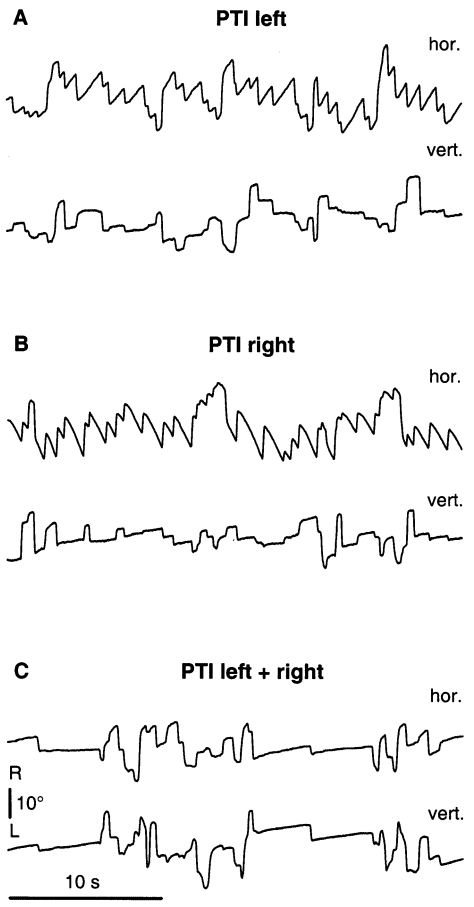

Fig. 4. Eye position in complete darkness in one cat during left, right, and bilateral inactivation of the pretectal region (PTI). Upper traces show horizontal, and lower traces show vertical eye movements. During left PTI (A), a horizontal spontaneous nystagmus developed with slow phases directed to the right. During right PTI (B), a horizontal spontaneous nystagmus developed with slow phases directed to the left. When both pretectal regions were inactivated (C), no horizontal spontaneous nystagmus could be seen. There was no effect of PTI on vertical eye position. Calibration bars in the lower left corner represent 10 s and 10°, and upward deflections of the traces correspond to rightward and upward eye movements.

The gain was always less than 0.3, indicating that cats were only poorly able to stabilize downward retinal slip.

The asymmetrical up- and downward OKN was further studied with different stimulus velocities (Fig. 6). In the upward direction, the gain increased with slower stimulus velocities. In the downward direction, it was difficult to see a clear velocity effect because of the low gain. The strong asymmetry was maintained at all velocities.

3.2.3. Muscimol injection

In Fig. 5A, C, E (triangles) the gain of the slow phase OKN after application of muscimol into the left pretectal area is displayed again for 12 different directions of stimulus movement. PTI resulted in an inability to perform OKN at horizontal stimulus movements directed towards the injected site. Thus, the gain was zero or negative, i.e. in the latter case, spontaneous eye movements were performed against the direction of

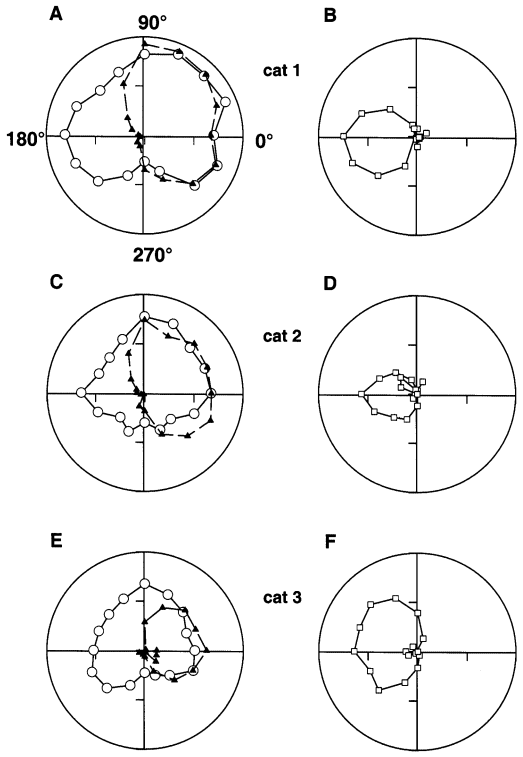

Fig. 5. Polar plots of slow phase gain of OKN during control and PTI of left pretectal region in three cats. In (A), (C), and (E), the gain is depicted for 16 directions of optokinetic stimulation at 12°/s during control (open circles) and PTI (triangles). (B), (D), and (F) give the difference of control and PTI gain, i.e. the gain loss by the inactivation. The radius of the outer circle in each polar plot indicates a gain of 1, i.e. when the eye-velocity equals stimulus velocity.

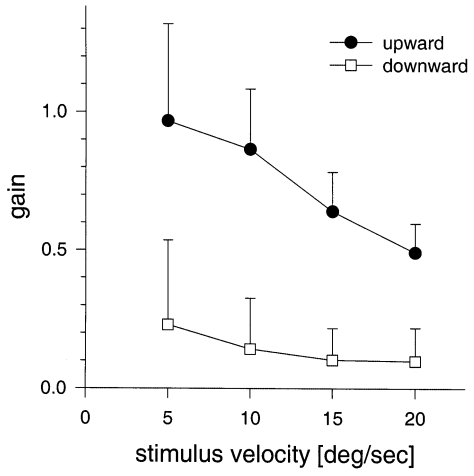

Fig. 6. Relationship of vertical slow phase gain to optokinetic stimulus velocity during bilateral PTI in one cat. The vertical bars show standard deviations for upward (circles) and downward (rectangles) optokinetic stimulus movements.

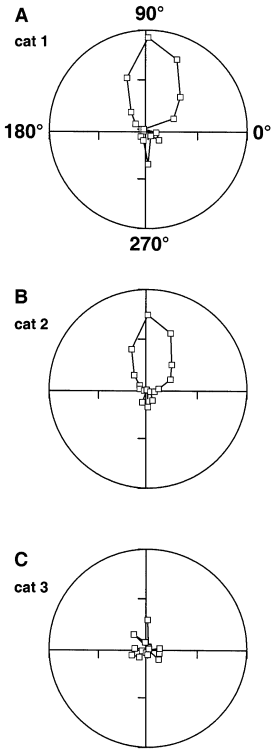

Fig. 7. Polar plots predicting the results of bilateral pretectal inactivation. For an explanation, see the main text. The conventions are the same as those in Fig. 7.

stimulus movement. However, we did not observe gains higher than 1 by an additive effect of spontaneous nystagmus in the direction towards the intact side. Neurons in the now inactivated NOT-DTN would have been suppressed by this stimulus direction anyhow. Thus, stimulus-driven horizontal components of OKN towards the intact side should be rather normal after PTI.

For Fig. 5B, D, F, the difference in gain between the controls and the PTI cases was calculated in each stimulus direction. These curves demonstrate the velocity signals that have been removed from the optokinetic reflex by left NOT-DTN inactivations. The curves look conspicuously like the neural tuning curve of a left NOT-DTN, as published by Hoffmann and Schoppmann (1981).

With unilateral inactivations, there was a complete elimination of ipsiversive horizontal nystagmic eye movements, including a loss of the ipsiversive horizontal component with oblique stimulus directions in cats 1 and 2 (Fig. 5B, D). Cat 3 (Fig. 5F) was also unable to perform an ipsiversive horizontal OKN. In addition, in this cat, upward and downward nystagmic eye movements were affected, but to a smaller extent than horizontal directions.

To predict the loss of nystagmic eye movements during bilateral pretectal inactivation, we calculated the mirror images of the gain losses in Fig. 5B, D, F and subtracted them from the control values. The results are shown in Fig. 7 for all three cats to reveal the variance between the animals. For cats 1 and 2, nystagmic eye movements should be possible only in a small range of upward stimulation, whereas the downward component would be very small. This pattern is extremely reduced in cat 3, which should perform an OKN only into the upward direction.

On average, our prediction of possible nystagmic eye movements after bilateral PTI resulted in a potential OKN into upward directions of a very narrow range of about $\pm 25°$. OKN into all other directions was predicted to be strongly reduced or impossible.

3.2.4. Bilateral pretectal inactivation

To test this prediction, a bilateral pretectal inactivation was performed in cat 2 (Fig. 8). During the control situation (Fig. 8A), the gain of optokinetic eye movements was roughly symmetrical around the horizontal direction but showed asymmetry in the vertical direction. When the left pretectal area was inactivated, no OKN to the left side could be performed (Fig. 8B). During right pretectal inactivation, the ability of optokinetic eye movements to the right was destroyed

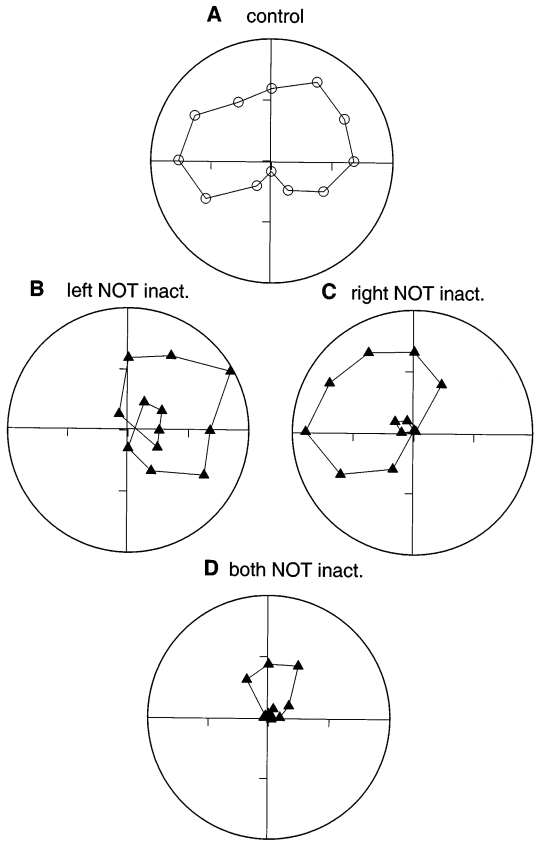

Fig. 8. Slow phase gain in cat 2 with bilateral pretectal inactivation, showing the gain during control (A), during left (B) and right (C) unilateral and during bilateral pretectal inactivation (D). The conventions are the same as those in Fig. 7.

(Fig. 8C). The unilateral PTI showed that during optokinetic stimulation in the direction of the lesioned NOT-DTN, slow-phase eye movements were directed in the opposite directions, i.e. towards the intact NOT-DTN. This is indicated by the triangles, which show a horizontal gain directed completely to the right during left PTI and to the left during right PTI.

When both left and right pretectal areas were inactivated (Fig. 8D), OKN could be performed with a low gain only in upward directions. Left-, right- and downward optokinetic movements were impossible during bilateral PTI. These results support our predictions of the loss of OKN made in Fig. 7.

4. Discussion

In the awake cat, NOT-DTN neurons showed increased activity in the light during the slow phase of OKN but also during periods of observing a stationary pattern when no OKN was visible. They showed best responses at stimulus velocities of about 5–20°/s and a strong directional selectivity for horizontal stimulation, which revealed their exclusive sensitivity to ipsiversive direction of retinal slip. The activity of NOT-DTN neurons during retinal slip, their directional selectivity, and the appearance of nystagmic eye movements during electrical stimulation of the NOT-DTN prove that these neurons are directly involved in gaze stabilisation and eliciting optokinetic nystagmus in all mammals studied so far.

These results correspond to behavioural experiments in that temporary inactivation of the left NOT-DTN with muscimol led to spontaneous nystagmus in the dark in the preferred direction of neurons of the intact NOT-DTN, i.e. to the right. An OKN toward the lesioned side was abolished. In contrast, with optokinetic stimulation to the side contralateral to the inactivation, cats showed normal OKN gain during binocular stimulation. Bilateral inactivation prevented OKN in both horizontal directions. Cats show a clear asymmetry of the vertical OKN. Upward stimulation was more effective, and this asymmetry was retained during bilateral inactivation.

In previous studies (Precht & Strata, 1980; Kato et al., 1986; Schiff et al., 1990), the NOT-DTN was inactivated by electrolytic lesions or injection of drugs that destroyed the cell bodies. This caused severe damage to the brain tissue and was irreversible. However, some effects of the lesion, e.g. the spontaneous nystagmus, subsided within several days (Schiff et al., 1990). This shows that there was plasticity in the optokinetic system to compensate for the deficits. With temporary lesions mediated by muscimol compensation was impossible, and thus, phenomena unaffected by the plasticity of the system could be observed. As revealed by control measurements, the effect of muscimol completely vanished 24 h after injection.

The time course of muscimol spread in brain tissue was investigated by Martin (1991). [3H]muscimol injected into the rat cortex had a spread of about 1.7 mm within the first 20 min postinjection, and the width remained relatively constant up to 120 min postinjection. After this time, diffusion of [3H]muscimol continued up to a radius of 3 mm. Assuming that the diffusion in the cat pretectal region is somehow similar to that in the rat cortex, we suppose that the diffusion of muscimol during the experimental session with a duration of at least 2 h never exceeded 2 mm. Thus, no structure involved in the generation of the vertical OKN could be inactivated because MTN and LTN in the cat were more than 2 mm lateral and caudal to the region that we injected. Consequently, the vertical eye movements were never affected by the inactivation. Also, a proposed influence of MTN/LTN on horizontal OKN (Clement & Magnin, 1984) cannot be affected by our inactivation.

Single-cell recordings in the NOT-DTN revealed that the preferred velocity was in the range of 5–20°/s. We tried to elicit OKN with retinal slip velocities in the same range to match the preferred velocity of NOT-DTN cells. From the used stimulus velocities and the measured eye movements, we could calculate retinal slip velocities in the range of 3–14°/s, which was well within the range of the preferred velocity of NOT-DTN neurons. For stimulation, we used a Julesz pattern that elicited optimal slow phases for velocities above 10°/s (Hamada, 1983; Maioli & Precht, 1984).

4.1. Vertical optokinetic nystagmus

In all cats, we observed a strong asymmetry of vertical OKN with upward gains reaching 60–70% higher values than downward gains. Such an asymmetry was also described by Precht (1981), King and Leigh (1982), Grasse and Cynader (1988), and Taillanter (1991). A similar asymmetry was observed in humans with upward OKN gains 10–20% higher than downward OKN gains (van den Berg & Collewijn, 1988; Murasugi & Howard, 1989; Ogino et al., 1996). In monkeys, an asymmetry in the opposite direction was observed: downward gain was 20–30% higher than upward gain (Takahashi & Igarashi, 1977; Matsuo & Cohen, 1984; Kato et al., 1986).

Because humans, monkeys and cats are frontal-eyed species, the optic flow of a textured ground during forward locomotion is in a downward direction and may elicit an optokinetic response. To suppress such undesired locomotion-induced OKN, species with a fovea have evolved a smooth pursuit system and the ability of fixation. However, this speculation is valid only for humans because in monkeys, the upward OKN

is more suppressed than the downward OKN. The functional reason for this asymmetry in monkeys is still unknown. In cats, the situation is more like that in humans. Since cats have no fovea, they must have evolved other mechanisms to suppress a locomotion-induced downward OKN. It seems that the much-reduced gain for downward stimulation is a functional adaptation to suppress downward OKN during locomotion.

4.2. Optokinetic nystagmus as a function of stimulus direction

Horizontal optokinetic responses in cats were studied previously with moving vertical stripes (Evinger & Fuchs, 1978; Donaghy, 1980), but smooth pursuit during slow-phase optokinetic eye movements was optimal over a large range of stimulus velocities when a random dot pattern was used (Maioli & Precht, 1984). Stimulating with a similar pattern, we could confirm the horizontal gain of 0.7–0.9 at stimulus velocities of 5–20°/s measured by these authors. For oblique gains, no comparable data are available. We could show that gains of 0.7–0.9 were also valid for oblique and upward directions.

Unilateral and bilateral pretectal lesions in the cat were previously performed by Precht and Strata (1980). In accordance with our study, they reported an absence of horizontal OKN towards the side of the lesion when one pretectal area was destroyed and a complete loss of horizontal OKN after bilateral pretectal lesions. These results are consistent with lesion studies in primates, which have shown that the horizontal slow phase of OKN towards the lesioned side was reduced in velocity or absent (Yakushin et al., 2000). In conclusion, the NOT-DTN has the same fundamental role in generating the slow phase of the horizontal OKN in different species.

4.3. Functional considerations

We studied the influence of the loss of horizontal OKN after unilateral NOT-DTN lesion onto optokinetic responses to oblique directions of stimulation. The horizontal components of the slow phases of OKN were completely absent, which resulted in an inability to perform OKN also in oblique directions. This loss of the horizontal components seems to be of equal amount for upward and downward oblique directions because the polar plots showing the magnitude of the gain that was affected by PTI (Fig. 5B, D, F) are nearly symmetrical around the horizontal axis. An even stronger evidence for the complete loss of the horizontal component of OKN is the bilateral inactivation during which upward OKN was only possible in a sector of $< 45°$ around vertical. Thus, the NOT-DTN in the cat seems to be involved not only in the generation of the horizontal OKN but also in the generation of the horizontal component of oblique OKN. The cause of the drop in gain of pure vertical OKN in some cases of NOT-DTN inactivations remains to be clarified.

The activity of direction selective NOT-DTN neurons has to be compared with the activity of the other NOT-DTN to the same stimulus direction to construct the difference signal between the activity of the left and right nucleus (Hoffmann, 1982). This difference signal depends strongly on the movement direction, i.e. it is maximal in horizontal directions, decreases in oblique directions, and is zero in vertical directions because NOT-DTN cells are not influenced by vertical movements. During unilateral PTI, the system is inbalanced, and no proper difference signal can be calculated. Thus, the horizontal component in the direction of the lesioned NOT-DTN is abolished, and not only the horizontal OKN but also oblique nystagmic eye movements are strongly reduced. During bilateral PTI, the driving force of both NOT-DTNs is absent, which results in the inability to perform OKN to horizontal and oblique stimulus directions.

Our prediction in Fig. 7 is based on the assumption of a linear summation of the outputs of both NOT-DTNs because we simply mirrored the gain loss of Fig. 5B, D, F and subtracted them from the control values. This resulted in the hypothesis that, during bilateral PTI, only upward OKN should be possible. Since we obtained exactly this effect during our bilateral PTI, we can conclude that the difference signal between the two NOT-DTNs is indeed calculated linearly.

It is interesting to note that the activity of LTN and MTN neurons does not add to horizontal gain components. We somehow expected this because their tuning curves are as broad as those of NOT-DTN neurons. The most plausible explanation to us is that the activity of the nuclei in the accessory optic system and NOT is not combined to create OKN in different directions before the eye muscles but stays largely separate for horizontal and vertical stabilizing eye movements. Thus, our experiments show that NOT-DTN provides the drive only for the horizontal eye muscles. After inactivation of the NOT-DTN output, no horizontal component of gaze stabilisation is left. Whether this specificity is also true with respect to LTN/MTN and vertical eye movements has to be shown with inactivations of these structures in further experiments.

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